

Amendments to the Claims

The listing of claims will replace all prior versions, and listings of claims in the application.

1. (Currently amended) A method for isolation of biological macromolecules, said method comprising contacting a filter with a biological sample comprising said biological macromolecules and cellular debris, wherein said filter comprises a first filter layer and a second filter layer wherein said first filter layer and said second filter layer are within the same hollow body, and such that said first filter layer is contacted with said biological macromolecules before said second filter layer, and wherein said first filter layer has a pore size smaller than said second filter layer such that said first filter layer retains cellular debris and said second filter layer allows said biological macromolecules to pass and said biological macromolecules are isolated.

2. (Original) The method of claim 1, wherein said biological sample is a cellular lysate.

3. (Original) The method of claim 2, wherein said cellular lysate is derived from eukaryotic cells.

4. (Original) The method of claim 2, wherein said cellular lysate is derived from prokaryotic cells.

5. (Original) The method of claim 3, wherein said eukaryotic cells are selected from the group consisting of fungi, fish cells, yeast cells, plant cells and animal cells.

6. (Original) The method of claim 1, wherein said biological macromolecules are nucleic acid molecules.

7. (Original) The method of claim 1, wherein said biological macromolecules are protein molecules.

8. (Original) The method of claim 6, wherein said nucleic acid molecules are RNA molecules.

9. (Original) The method of claim 8, wherein said RNA molecules are mRNA molecules.

10. (Original) The method of claim 6, wherein said nucleic acid molecules are DNA molecules.

11. (Original) The method of claim 10, wherein said DNA molecules are vectors or plasmids.

12.-15. (Canceled)

16. (Previously presented) The method of claim 1, wherein said pore size of said second filter layer is about 1 μm to 500 μm .

17. (Previously presented) The method of claim 16, wherein said pore size of said second filter layer is about 10 μm to 70 μm .

18. (Previously presented) The method of claim 17, wherein said pore size of said second filter layer is about 20 μm .

19.-20. (Canceled)

21. (Previously presented) The method of claim 1, wherein said first filter layer comprises pores of sufficient size to retard the flow of cellular debris and particles.

22. (Previously presented) The method of claim 21, wherein said pores of said first filter layer are about 0.1 μm to 1.0 μm in diameter.

23. (Previously presented) The method of claim 21, wherein said pores of said first filter layer are about 0.2 μm in diameter.

24. (Previously presented) The method of claim 1, wherein said second filter layer is comprised of glass fibers, silica, paper, cellulose, nitrocellulose, diatomaceous earth, and acetylated cellulose.

25. (Previously presented) The method of claim 1, wherein said first filter layer is comprised of one or more materials selected from the group consisting of hydrophobic polysulfone, hydrophilic polyether sulfone, cellulose, acetylated cellulose, nitrocellulose, polyester, polyolefin, scintered polyethylene, porous ceramics, silica, polypropylene, paper, and polysaccharide.

26. (Canceled)

27. (Previously presented) The method of claim 1, wherein said first filter layer has an average pore size of about 0.2 μm , and said second filter layer has an average pore size of about 20 μm .

28. (Previously presented) The method of claim 1, wherein said first filter layer is provided in a form selected from the group consisting of wafer, cylindrical, rectangular, beads, gels, square, cartridge, swab tip, plug, frit, membrane, sheets or inserts.

29. (Previously presented) The method of claim 1, wherein said filter is provided in a form that is suitable to be inserted into a tube, microspin tube, microfuge tube, spin cartridge, vial, ampule, bag or suitable to fit multi-well plates typically used in processing of multiple samples, including, 6-well plates, 12-well plates, 24-well plates, 48-well plates, 96-well plates, 384-well plates, and the like, or suitable to fit into other plate sizes such as 35 mm plates, 60 mm plates, 100 mm plates, or 150 mm plates, and the like.

30. (Original) The method of claim 1, wherein the flow of the sample is facilitated by centrifugation, gravity, pressure, vacuum, or any combination thereof.

31. (Currently amended) A method for isolation of biological macromolecules, said method comprising;

(a) contacting cells or cellular source containing the macromolecules of interest with a composition capable of lysing all or substantially all of said cells to give a lysate; and

(b) contacting the lysate with a filter, wherein said filter comprises two filter layers, with a first filter layer in contact with a second filter layer such that said first filter layer is contacted with said lysate before said second filter layer, and wherein said first filter layer has a pore size smaller than said second filter layer such that said first filter layer retains cellular debris and said second filter layer allows said biological macromolecules to pass; and

(c) promoting the flow through the filter thereby isolating biological macromolecules.

32.-54. (Canceled)

55. (Currently amended) A method for isolating biological macromolecules comprising, separating a lysed natural source in a sample by filtration, wherein said sample is passed through a filter comprising a first filter layer and a second filter layer wherein said first layer and said second layer are in contact with each other such that said first filter layer is contacted with said biological macromolecules before said second filter layer, and wherein said first filter layer has a pore size smaller than said second filter layer such that said first filter layer retains cellular debris and said second filter layer allows said biological macromolecules to pass and biological macromolecules are isolated.

56. (Previously presented) The method according to claim 55, wherein the flow through the filter is promoted by applying positive or negative pressure, or by gravity, or by gravity increased by centrifugation, or by a combination thereof.

57. (Previously presented) The method according to claim 55, wherein said biological macromolecule is a plasmid DNA or genomic DNA having a size of from 1 to 50 kb.

58.-60. (Canceled)

61. (Previously presented) The method according to claim 55, wherein said first filter layer has a pore size of 0.1 to 1.0 μm , and the second filter layer has a pore size of 1 to 500 μm .

62. (Previously presented) The method according to claim 55, wherein said first filter layer comprises one or more materials selected from the group consisting of hydrophobic polysulfone, hydrophilic polyether sulfone, cellulose, acetylated cellulose, nitrocellulose, polyester, polyolefin, scintered polyethylene, porous ceramics, silica, polypropylene, paper, and polysaccharide.

63. (Previously presented) The method according to claim 55, wherein said second filter layer is comprised of glass fibers, silica, paper, cellulose, nitrocellulose, diatomaceous earth, and acetylated cellulose.

64.-65. (Canceled)

66. (Previously presented) The method of any of claims 1, 31, or 55, wherein said second filter layer shears genomic DNA.

67. (Currently amended) A method for isolating DNA molecules from cell lysates, said method comprising contacting a filter with said lysate, wherein said filter comprises a multilayered filter bed comprising at least a first filter layer and a second filter layer, and wherein said first filter layer has a pore size smaller than said second filter layer such that said first filter layer retains cellular debris and the second filter layer comprises pores of sufficient size to shear DNA molecules and the second filter layer allows the DNA molecules to pass and DNA molecules are isolated.

68. (Previously presented) The method of claim 67, wherein said pores of said first filter layer are about 0.1 μm to 1.0 μm in diameter.

69. (Previously presented) The method of claim 67, wherein said pores of said first filter layer are about 0.2 μm in diameter.

70. (Previously presented) The method of claim 67, wherein said DNA molecules are plasmid DNA or genomic DNA having a size of from 1 to 50 kb.

71. (Currently amended) A method for isolating biological macromolecules from cell lysates, said method comprising contacting a filtration apparatus assembled into a cartridge housing with said lysate, wherein said filtration apparatus comprises at least a first filter on top of a second filter, wherein the first and second filters are secured with an insert, and wherein the first filter is contacted with said lysate before said second filter such that said first filter layer retains cellular debris and said second filter layer allows said biological macromolecules to pass and the biological macromolecules are isolated.